

HALOPHILIC OR HALOTOLERANT FUNGI OF FOUR SEEDS FROM EGYPT

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SUMMARY. — On 15% NaCl-water agar at 28°C, 5 genera and 19 species in addition to one variety were collected from soybean (5 genera and 18 species +1 variety), chick-pea (3 genera and 10 species +1 var.), lentil (4 genera and 11 species) and sesame seeds (2 genera and 5 species).

Aspergillus (14 species) and *Penicillium* (4 species) were the most common genera on the 4 types of seeds. From these genera *A. ochraceus*, *A. sydowi*, *A. amstelodami*, *A. niger*, *A. montevidensis*, *A. repens*, *P. chrysogenum* and *P. jenseni* were prevalent in 4 or 3 types of seeds.

RÉSUMÉ. — Sur milieu agar (NaCl 15%), 5 genres, 19 espèces et 1 variété de champignons ont été isolés sur graines de soja (5 genres et 18 espèces +1 variété), de pois chiche (3 genres et 10 espèces +1 var.), de lentille (4 genres et 11 espèces) et de sésame (2 genres et 5 espèces).

Les genres *Aspergillus* (14 espèces) et *Penicillium* (4 espèces) sont les plus communs sur les 4 types de graines. Parmi ces genres, *A. ochraceus*, *A. sydowi*, *A. amstelodami*, *A. niger*, *A. montevidensis*, *A. repens*, *P. chrysogenum* and *P. jenseni* sont prépondérants dans 3 ou 4 types de graines.

KEY WORDS : halophilic fungi, *Aspergillus*, *Penicillium*, Egypt.

INTRODUCTION

The term halophilic (or halotolerant) fungi is generally used to define fungi growing better on media containing high salt (sodium chloride) concentration. In Egypt, numerous investigations were carried out on the fungal flora of seeds and grains (ASSAWAH & ELAROSI, 1960; MOUBASHER & al., 1972; ADBEL-KADER & al., 1979; MOUBASHER & al., 1979; EL-KADY & al., 1982; MAZEN & al., 1984; EL-MAGHRABY, 1984), but none of the works have been focused on halophilic or halotolerant fungi. The present investigation was aimed to study composition, density and frequency of occurrence of halophilic or halotolerant fungi associated to soybean, chick-pea, lentil and sesame seeds.

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Table I : Numbers of cases of isolation (out of 25 samples) and total counts (calculated per 25 seeds in every sample) of halophilic fungi recovered from soybean, chick-pea, lentil and sesame seeds on 15 % NaCl - water agar at 28°C.

Tableau I : Genres et espèces de champignons isolés sur 4 types de graines.

Genera and Species	Soybean		Chick-pea		Lentil		Sesame	
	TC	NCI	TC	NCI	TC	NCI	TC	NCI
Total count	517		474		208		15	
<i>Aspergillus</i>	450	25	418	25	47	16	9	5
<i>A. amstelodami</i> (Mangin) Thom & Church	112	19	202	18	9	3	—	—
<i>A. ochraceus</i> Wilhelm	90	14	64	14	4	2	2	1
<i>A. sydowii</i> (Bain & Sart.) Thom & Church	82	14	78	11	22	11	6	4
<i>A. niger</i> Van Tieghem	46	9	9	4	3	2	—	—
<i>A. flavus</i> Link	33	7	7	6	—	—	—	—
<i>A. montevidensis</i> Talica & Mackinnon	33	4	12	3	3	2	—	—
<i>A. repens</i> De Bary	13	3	35	6	1	1	—	—
<i>A. sulphureus</i> (Fres.) Thom & Church	15	3	—	—	—	—	1	1
<i>A. terreus</i> Thom	11	3	—	—	2	1	—	—
<i>A. chevalieri</i> var. <i>intermedius</i> Thom & Raper	6	3	1	1	—	—	—	—
<i>A. melleus</i> Yukawa	5	2	—	—	—	—	—	—
<i>A. candidus</i> Link	2	1	—	—	—	—	—	—
<i>A. egyptiacus</i> Moubasher & Moustafa	1	1	10	5	—	—	—	—
<i>A. ruber</i> (Konig, Spieckermann & Bremer) Thom & Church	1	1	—	—	—	—	—	—
<i>A. nidulans</i> (Eidam) Wint.	—	—	—	—	3	2	—	—
<i>Penicillium</i>	59	12	89	13	151	21	6	4
<i>P. chrysogenum</i> Thom	35	8	85	13	92	15	5	3
<i>P. jensenii</i> Zaleski	21	7	4	2	59	11	1	1
<i>P. lanosum</i> Westling	2	1	—	—	—	—	—	—
<i>P. nigricans</i> (Bain.) Thom	1	1	—	—	—	—	—	—
<i>Cladosporium sphaerospermum</i> Penzig	6	4	—	—	2	2	—	—
<i>Acrophialophora</i> species	2	1	—	—	—	—	—	—
<i>Monocillium</i> species	1	1	2	1	8	2	—	—

Occurrence remarks :

High occurrence : more than 12 cases; moderate occurrence : between 6-12 cases; low occurrence : between 3-5 cases; rare occurrence : less than 3 cases; TC : total count; NCI : number of cases of isolation out of 25 samples.

MATERIALS AND METHODS

Twenty five samples of each of soybean, chick-pea, lentil and sesame seeds, 500 g each, were collected from the markets in Egypt.

Determination of seed-borne fungi :

The seed-plate method was used for estimation of seed-borne fungi. Five seeds were put on the surface of sterile 15 % sodium chloride-water agar + rose bengal (1/15000) as a bacteriostatic agent (SMITH & DAWSON, 1944). Five plates were used for each seed sample. The plates were incubated at 28°C for 15-20 days, during which the growing fungi were identified, counted and calculated per 25 seeds in every sample.

The colonies of slow growing fungi were transferred to slants (10 % NaCl-Czapek's agar medium) to ensure precise counting and then to plates for identification.

RESULTS AND DISCUSSION

The moisture content of the seeds tested was generally low and fluctuated between 4.1-5.6 %, 3.8-8.3 %, 2.3-13.8 % and 0.2-4.5 % in soybean, chick-pea, lentil and sesame seeds, respectively.

The best count of fungi was recorded on soybean (517 colonies per 25 seeds in every sample) followed by chick-pea (474 colonies), lentil (208 colonies) and sesame (15 colonies) seeds.

Nineteen halophilic species and 1 variety of *Aspergillus chevalieri* which belong to 3 genera in addition to two unidentified species of *Acrophialophora* and *Monocillium* were collected from soybean (5 genera and 18 species + 1 variety), chick-pea (3 genera and 10 species + 1 variety), lentil (4 genera and 11 species) and sesame seeds (2 genera and 5 species) as shown in Table 1. All of these fungi were firstly recorded as halophilic or halotolerant organisms from Egyptian seeds; but all of these fungi were recovered previously from Egyptian desert and cultivated soils on 5 %, 10 % and 15 % NaCl-Czapek's agar (MOUBASHER & al., 1985; ABOUL-NASR, 1981) as well as from Saudi Arabian desert soils on 5 NaCl-Czapek's agar (ABDEL-HAFEZ, 1981).

Aspergillus was the most common genus on soybean, chick-pea and sesame seeds, but retreated to the second place on lentil. It occurred in 100 %, 100 %, 64 % and 20 % of the samples contributing 87 %, 88.2 %, 22.6 % and 60 % of total fungi on soybean, chick-pea, lentil and sesame seeds, respectively. It was represented by 14 species + 1 variety in all types of seeds (13 + 1 var., 8 + 1 var., 8 and 3 species on the four substrates, respectively) of which *A. ochraceus* and *A. sydowi* were common on the four types of seeds. These two species were isolated but with variable densities and frequencies, from soils in Egypt (MOUBASHER & al., 1985; ABOUL-NASR, 1981) and Saudi Arabia (ABDEL-HAFEZ, 1981) on 5-15 % NaCl-Czapek's agar. *A. amstelodami* was isolated in high

frequency of occurrence from soybean and chick-pea seeds (76 % and 72 % of the samples; 24.9 % and 48.3 % of total *Aspergillus*; 21.7 % and 42.6 % of total fungi, respectively), but it was less frequent in lentil seeds and completely absent in sesame. *A. niger*, *A. flavus*, *A. montevidensis*, *A. repens*, *A. sulphureus*, *A. terreus*, *A. chevalieri* var. *intermedius* and *A. egyptiacus* were recovered in moderate or low frequency on one or two substrates. The remaining *Aspergillus* species were less frequent and listed in Table 1. ABDEL-HAFEZ (1981) isolated 20 species of *Aspergillus* from desert soils in Saudi Arabia on 5 % NaCl-Czapek's agar, of which *A. amstelodami*, *A. chevalieri*, *A. ruber* and *A. ochraceus* were the most common. It is worthy mentioning that members of *Aspergillus glaucus* group (such as *A. amstelodami*, *A. chevalieri*, *A. montevidensis*, *A. ruber*, *A. repens* and *A. tonophilus*) of halophilic or halotolerant and osmophilic or osmotolerant nature (RAPER & FENNELL, 1965; MOUSTAFA & AL-MUSALLAM, 1975; ABDEL-HAFEZ & al., 1977; ABDEL-HAFEZ, 1981, 1982; ABOUL-NASR, 1981; MOUBASHER & al., 1985).

Penicillium occupied the second place with regard to the number of cases of isolation in soybean, chick-pea, and sesame seeds, and promoted to the first place in lentil. It occurred in 16 % - 84 % of the samples constituting 11.4 - 72.6 % of total fungi in the 4 types of seeds. It was represented by 4 species of which *P. chrysogenum* was the most common in all seeds tested. *P. jensenii*, *P. lanosum* and *P. nigricans* were less frequent. ABDEL-HAFEZ (1981), ABOUL-NASR (1981) and MOUBASHER & al. (1985) found that *P. notatum* (= *P. chrysogenum*) was the most prevalent *Penicillium* species in soils from Egypt and Saudi Arabia on 5-15 % NaCl - Czapek's agar medium.

Cladosporium sphaerospermum was isolated in low and rare frequencies of occurrence from soybean and lentil seeds (16 % and 8 % of the samples and 1.2 % and 1 % of total fungi, respectively), but it was completely absent in chick-pea and sesame seeds. This species was recovered, but with variable density and frequency, from Egyptian and Saudi Arabian soils on 5-15 % NaCl-Czapek's agar (ABDEL-HAFEZ, 1981; ABOUL-NASR, 1981; MOUBASHER & al., 1985). BAYLIS ELLIOT (1930) found that *Hormodendron* (= *Cladosporium*) was dominant in Dovey salt marshes.

Acrophialophora sp. and *Monocillium* sp. were isolated in rare frequency of occurrence from one and three substrates, respectively (Table 1). The preceding two genera were reported by ABOUL-NASR (1981) as highly osmophilic and halophilic ability.

In conclusion, there are basic similarities between fungi associated with the 4 types of seeds, but some species increased or decreased their frequencies of occurrence as well as total counts; the great majority of species recovered from Egyptian seeds are of halophilic (or halotolerant) nature; and there are no fungal flora characteristic of Egyptian soybean, chick-pea, lentil and sesame seeds when compared with results obtained from other types of seeds.

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